

## Effects of *Lactobacillus*-probiotic on the Performance and Immune Response of Chickens

Y. W. Ho, D. D. Vickneswary, N. Abdullah, I. Zulkifli and M. Zamri Saad.

Institute of Bioscience  
Universiti Putra Malaysia  
43400 UPM, Serdang, Selangor  
Malaysia

**Key words:** Probiotic, broilers, *Lactobacillus*, immune response, growth performance

### Introduction

During the last two decades, as awareness on the dangers of using sub-therapeutic levels of antibiotics as growth promoters in animals increased, probiotics, which include *Lactobacillus* cultures, have been considered as a substitute for antibiotic growth promoters in animal production. The probiotics, which upon ingestion, contribute some beneficial functions in the intestinal tract of the animal and exert health effects beyond inherent basic nutrition. Our earlier studies have shown that broiler chickens fed *Lactobacillus*-probiotic have better growth rate and feed conversion ratio, lower mortality rate, less pathogenic bacteria such as *E. coli* and *Salmonella* in their gastrointestinal tract, less noxious bacterial enzymes such as  $\beta$ -glucuronidase and  $\beta$ -glucosidase in their intestine, less body fat and less cholesterol than chickens fed without probiotic (Jin *et al.*, 1997, 1998a, b, 2000). Probiotics have also been found to exert an immunomodulatory effect on the host. Supplementing *Lactobacillus* cultures to the diets of newly hatched chicks and poults increased the production of anti-salmonella antibodies and function of T-cells (Durham *et al.*, 1993). The present study was conducted to determine the effects of *Lactobacillus*-probiotic on the performance and immune response of broilers.

### Materials and Methods

The *Lactobacillus* strains used and the method of preparation of the strains as a probiotic feed supplement were the same as those described by Jin *et al.* (1996, 1998a).

Four trials were carried out: (i) with chicks not challenged with enteropathogens, (ii) with chicks challenged with *Salmonella enteritidis*, (iii) with chicks challenged with *E. coli* and (iv) with chicks not challenged with enteropathogens and subjected to heat stress conditions. For the first trial, 270 1-day-old male broiler chicks (Avian 43) were randomly assigned to 18 cages of 15 chicks each. The chicks were divided into 3 dietary treatment groups (6 cages per treatment). The diets were: (i) a basal diet (control), (ii) a basal diet + 0.1% *Lactobacillus*-probiotic, and (iii) a basal diet + 50 mg oxytetracycline/kg feed. For the second and third trials, 200 1-day-old broiler chicks (Avian 43) were randomly assigned to 4 SPF cages for each trial. Each cage was assigned to one of the 4 treatments: (i) a basal diet (control), (ii) a basal diet and challenged with *S. enteritidis* or *E. coli* after 14 days (control challenged with *S. enteritidis* or *E. coli*), (iii) a basal diet + 0.1% *Lactobacillus*-probiotic, and (iv) a basal diet + 0.1% *Lactobacillus*-probiotic and challenged with *S. enteritidis* or *E. coli* after 14 days. The experimental period for all the 3 trials was 42 days. The chicks were weighed weekly and mortality was recorded as it occurred. At 21, 28, 35, and 42 days of age, the chicks were sacrificed. The blood and intestinal scrapings were collected. The spleen, liver and bursa were removed and weighed. Enzyme linked immunosorbent assay (ELISA) and resultant colour absorbance at 405 nm ( $A_{405}$ ) were used to measure the immunoglobulin (IgA and IgG) levels in serum and supernatant of the intestinal scrapings. For the fourth trial, 180 Shaver and 180 Hubbard 1-day-old female broiler chicks were assigned to 36 cages of 10 birds each. The diets were the same as those of the first trial. Live Newcastle disease vaccine was administered at 7 and 21 days of age. From 21 to 42 days of age, the chicks were subjected to 3-h episodes of heat stress at 36°C. Blood samples were analysed for antibody titres against Newcastle disease using ELISA.

### Results and Discussion

The results of the first trial showed that the feed conversion ratio of broilers fed *Lactobacillus*-probiotic (1.72) was significantly better than that of the control broilers (2.06) or broilers fed antibiotic (1.93) from 1 to 42 days of age. The body weight gain of probiotic-fed chickens (1950 g) was comparable to that of the antibiotic-fed (1922 g), but significantly more than that of the control (1690 g) chickens. Mortality rate was also significantly lower in chickens fed *Lactobacillus*-probiotic (2.57 %) when compared to the control (12.51 %) or antibiotic-fed chickens. There was no significant difference in organ weights among the chickens fed the 3 dietary treatments at 42 days of age. This showed that there was no inflammatory response by the chickens to the *Lactobacillus*-probiotic. Thus, using *Lactobacillus*-probiotic as an immunomodulator has an advantage over the other immunomodulators as it did not produce splenomegaly or hepatomegaly, which are very common effects of other immunomodulators (Yasutake *et al.*, 1984). At 42 days of age, the serum and intestinal IgA levels in chickens fed *Lactobacillus*-probiotic were significantly higher than those in the control or antibiotic-fed chickens. This indicated that the *Lactobacillus*-probiotic could enhance the serum and mucosal intestinal immunity of chickens. An important feature of immunological defence at the mucosal surface is the predominance of IgA. Symbiotic and indigenous microflorae contribute to the host's defence by increasing the number of IgA plasma cells in the mucosal surface of the intestinal wall. There was no significant difference in the serum and intestinal IgG levels among chickens fed the 3 dietary treatments. This indicated the absence of inflammatory processes.

The results of the second and third trials showed that the weight gains of probiotic-fed chickens with or without challenge with *S. enteritidis* or *E. coli* were significantly more than those of the control chickens with or without challenge with *S. enteritidis* or *E. coli* from 1 to 42 days of age. Mortality rate was the lowest in probiotic-fed chickens without challenge, followed by probiotic-fed chickens challenged with *S. enteritidis* or *E. coli*, and the highest in control chickens challenged with *S. enteritidis*. The relative weights of the liver and spleen of probiotic-fed chickens challenged with *S. enteritidis* or *E. coli* were not significantly different from those of the control or probiotic-fed chickens without challenge with the enteropathogens at 21, 28, 35 and 42 days of age. However, the relative weights of the liver and spleen of the control chickens challenged with *S. enteritidis* or *E. coli* were more than those of chickens in the other treatments, which indicated infection in the control chickens challenged with the enteropathogens. In the first week after challenge with *S. enteritidis* or *E. coli* (21 days of age), the serum and intestinal IgG levels of chickens fed *Lactobacillus*-probiotic were significantly higher than those of the control chickens (challenged with the enteropathogens), but at the end of the experimental period (42 days of age), the serum and intestinal IgG levels of the probiotic-fed chickens were significantly lower than those of the control chickens. Like IgG, the serum and intestinal IgA levels of chickens fed *Lactobacillus*-probiotic were also significantly higher than those of the control chickens in the first week after challenged with *S. enteritidis* or *E. coli* (21 days of age), and at the end of the experimental period (42 days of age), the serum and intestinal IgA levels of probiotic-fed chickens challenged with *S. enteritidis* or *E. coli* were significantly lower than those of the control chickens challenged with the enteropathogens. These results showed that the elimination of pathogenic *S. enteritidis* or *E. coli* was rapid in probiotic-fed chickens due to increased IgA serum and intestinal antibody levels. The higher IgA antibody level in the control chickens at the end of the experimental period indicated that the elimination of the enteropathogens was slow and the pathogens still persisted in the guts of the control chickens. In the fourth trial, after 3 weeks of heat exposure, chickens fed *Lactobacillus*-probiotic had significantly better weight gain and feed efficiency than the control chickens or those fed antibiotic. Hubbard chickens fed *Lactobacillus*-probiotic also had significantly higher antibody production against Newcastle disease than the control chickens at 42 days of age.

### Conclusions

The results of the study showed that *Lactobacillus*-probiotic could enhance the growth performance, feed efficiency and immune response of broiler chickens.

### Benefits from the study

The *Lactobacillus*-probiotic could be used as an alternative to antibiotics as a growth promoter. It could also be used as an oral adjuvant to prevent enteric infections. Production cost of chickens is lowered as growth performance and feed efficiency of the birds are enhanced.

### Literature cited in the text

- Dunham, H. J., Williams, C., Edens, F. W., Casas, I. A. and Dobrogosz, W. J. 1993. *Lactobacillus reuteri* immunomodulation of stressor-associated diseases in newly hatched chickens and turkeys. *Poult. Sci.* 72: 103.
- Jin, L. Z., Ho, Y. W., Abdullah, N. and Jalaludin, S. 1997. Probiotics in poultry: modes of action. *World's Poult. Sci. J.* 53: 351-368.
- Jin, L. Z., Ho, Y. W., Abdullah, N. and Jalaludin, S. 1998a. Growth performance, intestinal microbial populations and serum cholesterol of broiler diets containing *Lactobacillus* cultures. *Poult. Sci.* 77: 1259-1265.
- Jin, L. Z., Ho, Y. W., Abdullah, N., Ali, M. A. and Jalaludin, S. 1998b. Effects of adherent *Lactobacillus* cultures on growth, weight of organs and intestinal microflora and volatile fatty acids in broilers. *Anim. Feed Sci. Technol.* 70: 197-209.
- Jin, L. Z., Ho, Y. W., Abdullah, N. and Jalaludin, S. 2000. Digestive and bacterial enzyme activities in broilers fed diets supplemented with *Lactobacillus* cultures. *Poult. Sci.* 79: 886-891.
- Jin, L. Z., Ho, Y. W., Ali, M. A., Abdullah, N., Ong, B. K. and Jalaludin, S. 1996. Adhesion of *Lactobacillus* isolates to intestinal epithelial cells of chicken. *Lett. Appl. Microbiol.* 22: 229-232.
- Yasutake, N., Onwaki, M., Yokokura, T. and Mutai, M. 1984. Comparison of antitumor activity of *Lactobacillus casei* with other bacterial immunopotentiators. *Med. Microbiol. Immunol.* 173: 113-125.

### Project Publications in Refereed Journals

- Zulkifli, I., Abdullah, N., Mohd. Azrin, N. and Ho, Y. W. 2000. Growth performance and immune response of two commercial broiler strains fed diets containing *Lactobacillus* cultures and oxytetracycline under heat stress conditions. *Br. Poult. Sci.* 41: 593-597.

### Project Publications in Conference Proceedings

- Vickneswary, D. D., Ho, Y. W., Abdullah, N. and Zamri Saad, M. 2000. Growth performance and serum immunoglobulin (IgA) level of broilers fed diets containing *Lactobacillus*-probiotic. *Proceedings of the 22<sup>nd</sup> Malaysian Society of Animal Production Annual Conference on "Good Animal Husbandry Practices – the Future for Sustainable Food Production"*, pp. 215-216.

Vickneswary, D. D., Ho, Y. W., Abdullah, N. and Zamri Saad, M. 2000. Immunoglobulin (IgA and IgG) levels in intestinal scrapings of broilers fed diets containing *Lactobacillus*-probiotic. Proceedings of the 23<sup>rd</sup> Symposium of the Malaysian Society for Microbiology on "Microbes and Biotechnology for the Advancement of Health, Food Production and Agriculture", pp. 107-108.

Vickneswary, D. D., Ho, Y. W., Abdullah, N. and Zamri Saad, M. 2001. Effects of *Lactobacillus*-probiotic supplementation on growth performance of broilers challenged with *Salmonella enteritidis*. Proceedings of the 23<sup>rd</sup> Malaysian Society of Animal Production Annual Conference on "Globalisation and Livestock Production in Developing Nations", pp. 106-107.

<i>Expertise Development</i>					
Name of Graduate		Degree Awarded	Field of Expertise	Research Topic	Graduation Year
Vickeswary Dharmadas	David	MSc	Microbiology	Effects of <i>Lactobacillus</i> -probiotic on the Performance and Immune Response of Chickens	On-going

IRPA Project number 01-02-04-0455

UPM Research Cluster :AFF

Project Leader Ho Yin Wan